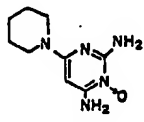




BG

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁴ : A61K 7/06, 9/66, 31/685 A61K 37/22, B01J 13/02	A1	(11) International Publication Number: WO 88/ 07362 (43) International Publication Date: 6 October 1988 (06.10.88)
(21) International Application Number: PCT/US88/00995 (22) International Filing Date: 28 March 1988 (28.03.88) (31) Priority Application Number: 032,512 (32) Priority Date: 30 March 1987 (30.03.87) (33) Priority Country: US (71) Applicant: LIPOSOME TECHNOLOGY, INC. [US/US]; 1050 Hamilton Court, Menlo Park, CA 94025 (US). (72) Inventors: USTER, Paul, Steven ; 883 Warren Way, Palo Alto, CA 94303 (US). QUINN, Yolanda, Platon ; 631 Larchmont Drive, Daly City, CA 94015 (US). (74) Agents: DEHLINGER, Peter, J. et al.; Ciotti & Murashige, Irell & Manella, 545 Middlefield Road, Suite 200, Menlo Park, CA 94025 (US).	(81) Designated States: AT (European patent), AU, BE (European patent), CH (European patent), DE (European patent), DK, FR (European patent), GB (European patent), IT (European patent), JP, LU (European patent), NL (European patent), NO, SE (European patent). Published <i>With international search report.</i>	
(54) Title: NON-CRYSTALLINE MINOXIDIL COMPOSITION AND METHOD OF APPLICATION		
(57) Abstract		
<p>An aqueous, non-crystalline minoxidil composition for topical use. The composition contains minoxidil complexed with an amphipathic compound having a pK less than about 5 and containing a single lipophilic chain moiety and a sulfate, sulfonate, phosphate and phosphonate polar moiety. The composition may be formulated in ointment form, in an aqueous vehicle, or dispersed in a fluorochlorocarbon solvent, for spray delivery from a self-propelled spray device. The minoxidil in the composition remains in non-crystalline form for a period of at least several hours after application to the skin, thus overcoming the prior art problem of the minoxidil tending to revert to an insoluble crystalline form when prior art compositions are applied to the skin.</p>	<div style="display: flex; flex-direction: column; align-items: center;"> <div style="display: flex; justify-content: space-around; width: 100%;"> <div style="text-align: center;">  <p>a. MINOXIDIL</p> </div> <div style="text-align: center;"> $\text{CH}_3-(\text{CH}_2)_{10}-\text{CH}_2-\text{O}-\text{C}(=\text{O})-\text{CH}(\text{SO}_3\text{Na})-\text{CH}_2-\text{C}(=\text{O})-\text{ONa}$ <p>b. LAURETH SULFOSUCCINATE HEMI-ESTER</p> </div> </div> <div style="display: flex; justify-content: space-around; width: 100%;"> <div style="text-align: center;"> $\text{CH}_3-(\text{CH}_2)_{15}-\text{CH}_2-\text{O}-\text{C}(=\text{O})-\text{CH}(\text{SO}_3\text{Na})-\text{CH}_2-\text{C}(=\text{O})-\text{ONa}$ <p>c. LAURYL SULFOSUCCINATE HEMI-ESTER</p> </div> <div style="text-align: center;"> $\begin{array}{c} \text{OH} \\ \\ \text{CH}_2-\text{CH}_2-\text{CH}_2-\text{O}-\text{PO}_3\text{H}_2 \\ \\ \text{O} \\ \\ \text{C}=\text{O} \\ \\ (\text{CH}_2)_n \\ \\ \text{CH}_3 \end{array}$ <p>d. LYSO PHOSPHATIDIC ACID</p> </div> </div> <div style="text-align: center; margin: 10px 0;"> $\text{CH}_3-(\text{CH}_2)_7-\text{CH}=\text{CH}(\text{CH}_2)_7-\text{CH}_2\text{OCH}_2\text{CH}_2)_3-\text{OPO}_3\text{H}_2$ <p>e. CRODAFOS N3 ACID (OLETH 3 PHOSPHORIC ACID)</p> </div> <div style="display: flex; justify-content: space-around; width: 100%;"> <div style="text-align: center;"> $\text{CH}_3(\text{CH}_2)_n-\text{PO}_3\text{Na}_2$ <p>f. ALKYL PHOSPHONATE</p> </div> <div style="text-align: center;"> $\text{CH}_3(\text{CH}_2)_n-\text{OSO}_3\text{Na}$ <p>g. ALKYL SULFATE ESTER</p> </div> </div> <div style="display: flex; justify-content: space-around; width: 100%;"> <div style="text-align: center;"> $\text{CH}_3-(\text{CH}_2)_n-\text{SO}_3\text{Na}$ <p>h. ALKYL SULFONATE</p> </div> <div style="text-align: center;"> $\begin{array}{c} \text{CH}_3(\text{CH}_2)_7-\text{CH}=\text{CH}(\text{CH}_2)_7-\text{C}(=\text{O}) \\ \\ \text{NH} \\ \\ \text{NaO}-\text{C}(=\text{O})-\text{CH}_2-\text{CH}-\text{C}(=\text{O})-(\text{OCH}_2\text{CH}_2)_3 \\ \\ \text{SO}_3\text{Na} \end{array}$ <p>i. OLEAMIDO POLYETHYLENE GLYCOL-3 SULFOSUCCINATE HEMI-ESTER</p> </div> </div> </div>	

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AT	Austria	FR	France	ML	Mali
AU	Australia	GA	Gabon	MR	Mauritania
BB	Barbados	GB	United Kingdom	MW	Malawi
BE	Belgium	HU	Hungary	NL	Netherlands
BG	Bulgaria	IT	Italy	NO	Norway
BJ	Benin	JP	Japan	RO	Romania
BR	Brazil	KP	Democratic People's Republic of Korea	SD	Sudan
CF	Central African Republic	KR	Republic of Korea	SE	Sweden
CG	Congo	LI	Liechtenstein	SN	Senegal
CH	Switzerland	LK	Sri Lanka	SU	Soviet Union
CM	Cameroon	LU	Luxembourg	TD	Chad
DE	Germany, Federal Republic of	MC	Monaco	TG	Togo
DK	Denmark	MG	Madagascar	US	United States of America
FI	Finland				

NON-CRYSTALLINE MINOXIDIL COMPOSITION AND METHOD OF APPLICATION

1. Field of the Invention

10 The present invention relates to a topical minoxidil composition, and in particular, to a non-crystalline composition which can be applied in spray or ointment form.

2. Background of the Invention

15 Minoxidil is an arterial dilator which has been used, in oral form, in the treatment of hypertension to lower blood pressure. More recently, the drug has been shown to stimulate new hair growth, when applied topically, in cases of male pattern
20 baldness. Initial clinical studies with a topical form of the drug indicate that reversal of male pattern baldness is most favorable in younger men, and where recent hair loss has occurred, but that new hair growth is observed in a significant percentage of older men
25 and/or where in cases of long-term baldness.

 The drug itself is a piperidiny pyrimidine compound which is poorly soluble in water and in most water-immiscible organic solvents such as chloroform. Heretofore, minoxidil has been formulated, for topical
30 use, in an ethanol-based ointment vehicle containing ethanol, propylene glycol and water. The solubility of the drug in pure propylene glycol is between about 7-9% by weight, and in an ethanol/propylene glycol/water vehicle, only about 2%. On drawback of the formulation

is the tendency of the minoxidil to revert to an insoluble crystalline form when applied to the skin, as the ethanol solvent evaporates. Whether due to the tendency of the drug to crystallize or other factors, the minoxidil formulation shows relatively inefficient uptake by the skin. Another limitation is the limited solubility of the drug in the ethanol/propylene glycol/water vehicle. Further, evaporation of ethanol, when the formulation is applied to the skin, leaves a viscous propylene glycol/water residue which may be objectionable to many users. The drug is poorly soluble in water and practically insoluble in lipophilic solvents, such as chloroform. Therefore, predominantly water-based or propellant-solvent formulations have not been feasible heretofore.

European patent application No. 177,223 discloses a liposomal minoxidil composition in which minoxidil is present (1) in solution form possibly in a supersaturated state, either encapsulated in lipid vesicles, or in the aqueous or lipid phases of a liposome suspension, and (2) in a finely divided crystalline (solid) form both within and outside the lipid vesicles. Preferred embodiments of the composition are formed by hydrating a minoxidil lipid film containing a saturated phosphatidylcholine (PC), such as dipalmitoylphosphatidylcholine (DPPC), cholesterol, and minoxidil with an aqueous solution containing minoxidil in ethanol/propylene glycol and water. Minoxidil is present at a final weight concentration of between about 1.2-3%. The composition was found to contain liposomes of various sizes between about 1 μ to 15 μ , and minoxidil crystals. Although the formulation is reported to increase drug uptake by the epidermis when applied topically to skin, it has the

same limitation as the above non-liposomal formulation in that the drug is applied to the skin largely in crystalline form.

5 3. Summary of the Invention

It is one general object of the present invention to provide a non-crystalline minoxidil composition that can be formulated in a water-based or lipophilic-solvent vehicle.

10 Another object of the invention is to provide such a formulation which gives enhanced transdermal penetration of the drug.

The invention includes a non-crystalline minoxidil composition in which minoxidil is complexed with an amphipathic compound having a pK of less than about 5, and containing a single lipophilic chain and a polar head moiety selected from a sulfate, sulfonate, phosphate, or phosphonate free acid. The molar ratio of the amphipath to minoxidil is at least about 1:1, and the composition has a preferred pH between about 4-6. Preferred amphipathic compounds include sulfosuccinic acid hemiesters and alkyl phosphonates and phosphate esters, and exemplary sulfosuccinic acid hemiesters include ethoxylated sulfosuccinic acid hemiesters, such as the free acids of laureth, lauryl or oleamido-polyethylene glycol sulfosuccinate.

25 In one embodiment, the composition is formulated to include vesicle-forming lipids, such as PC, to form a minoxidil/liposomal suspension. The vesicle-forming lipids are preferably included at a molar ratio of between about 1:1 and 4:1 lipid to minoxidil.

30 In another embodiment, the composition is formulated to include surfactant components, to form an

emulsion or stable microemulsion/minoxidil composition. The minoxidil/amphipath complex may also be formulated in a micellar dispersion.

The minoxidil composition may be administered
5 in either spray or ointment form. A convenient spray formulation includes the minoxidil/ester complex dispersed in a chlorofluorocarbon propellant solvent. Phospholipid, such as PC, may be added to achieve a molecular monodisperse form of the drug in the solvent.
10 The minoxidil spray is directed against the topical area of interest, to deposit (with solvent evaporation) a noncrystalline minoxidil composition on the skin.

In a cream or ointment formulation, the minoxidil composition is dispersed in non-crystalline
15 form in an aqueous medium, in the presence of absence of suspended liposomal or lipid-emulsion particles. The formulation typically contains 1-6% minoxidil, at least an equal molar amount of the amphipathic compound, and between about 60-90% water. In a liposomal formulation,
20 the vesicle-forming lipids are present at a weight ratio of lipids to minoxidil of between about 1:1 and 4:1, where the transdermal uptake of minoxidil is reduced at higher lipid to drug ratios.

More generally, this composition includes a
25 noncrystalline minoxidil composition in which minoxidil is (a) present at a weight concentration of between about 1-6%; (b) complexed with an amphipathic compound having a pK less than about 5, at an amphipath:minoxidil molar ratio of at least about 1:1; (c) dispersed in an
30 aqueous or lipophilic-solvent carrier; and (d) maintained in noncrystalline form for a period of at least several hours after application to the skin. The amphipathic compound is preferably one which promotes transdermal uptake of the drug.

Also forming part of the invention is a method for applying minoxidil topically in a substantially noncrystalline form which remains in non-crystalline form at least several hours after application to the skin. The method includes complexing the minoxidil with an amphipathic compound having a pK less than about 5, and containing a single lipophilic chain moiety and a polar head moiety selected from a sulfate, sulfonate, phosphate, or phosphonate free acid. Specific formulations, and methods of application of the complex to the skin are as described above.

These and other objects and features of the invention will become more fully apparent when the following detailed description of the invention is read in conjunction with the accompanying drawings.

Brief Description of the Drawings

Figure 1 shows the molecular structure of minoxidil (1a) and the structures of several amphipathic compounds which promote solubilization of minoxidil in aqueous and lipophilic solvents and lipid bodies, and enhance transdermal uptake of the drug (1b-1h);

Figure 2 shows the molecular structures of several amphipathic compounds which promote solubilization of minoxidil uptake, but do not enhance skin penetration of the drug;

Figure 3 is a plot showing changes in minoxidil solubility, in a 20% laureth sulfosuccinate mixture, as a function of pH;

Figure 4 is a plot showing the increase in minoxidil solubility with increased concentrations of laureth sulfosuccinate in a pH 5.0 mixture;

Figure 5 shows transdermal uptake curves, over a 24 hour period, for control (open squares), 5%

minoxidil/sulfosuccinate (open diamonds), 2% minoxidil/sulfosuccinate (crosses), and 2% minoxidil/taurocholate (open triangles) compositions;

Figure 6 shows transdermal uptake curves for control (open squares) and a 1% minoxidil/phosphate monoester composition (crosses);

Figure 7 shows transdermal uptake curves for lauryl sulfosuccinate/PC liposome compositions containing either 2% (crosses) or 5% (open diamonds) minoxidil, a 1% minoxidil/cholesterol sulfate composition (open triangles), and the control composition (open squares);

Figure 8 shows transdermal uptake curves for a 2% minoxidil/oleamido-PEG-2/PC liposome formulation (open diamonds), 2% lauryl sulfosuccinate/PC liposomes (crosses), and the control formulation (open squares); and

Figure 9 shows transdermal uptake curves for a 2% minoxidil/lysoPA/PA composition (crosses), a 2% minoxidil/PA composition (open diamonds), and the control formulation (open squares).

Detailed Description of the Invention

I. Minoxidil Composition in Aqueous Medium

The minoxidil composition of the invention is formed by complexing minoxidil with the free acid of an amphipathic compound. The amphipathic compound acts to solubilize minoxidil in both aqueous and lipophilic solvents, and preferred compounds also enhance transdermal uptake of the drug. This section describes amphipathic compounds for use in the invention, and methods of preparing a water-soluble minoxidil

composition which is suitable for ointment and cream formulations.

A. Amphipathic Compounds

5 Figure 1 shows the molecular structure of minoxidil (1a). The piperidiny1 pyrimidinediamine compound is relatively soluble in alcohol, but poorly soluble in water (about 0.2-0.3%), and practically insoluble in chloroform.

10 The figure also shows the molecular structure of several specific and general types of amphipathic compounds which promote solubilization of the drug in both aqueous and lipophilic solvents and, according to an important feature of the invention, also enhance
15 transdermal uptake of the drug. The amphipathic compounds in this class are characterized by (a) a single lipophilic chain moiety and a polar head group moiety selected from a sulfate, sulfonate, phosphate, or phosphonate free acid, where the pK of the compound is
20 no greater than about 5.0. The exemplary compounds shown in the figure include laureth sulfosuccinate hemiester (1b) and lauryl sulfosuccinate (1c), representative of sulfosuccinate hemiesters, lysophosphatidic acid (1d) and monoalkyl phosphate
25 esters, such as Crodafos™ N3 (1e), monoalkyl phosphonates (1f), monoalkyl sulfate esters, (1g), monoalkyl sulfonates (1h), and oleamido-PEG-2-sulfosuccinate (1i), representative of amido-linked amphipaths.

30 As can be appreciated from the compounds shown in Figure 1, the lipophilic chain moiety may be a pure hydrocarbon chain, or may contain ether or other chain linkages, such as internal ester or amide linkages. The chains lipophilic chains preferably include at least

about 6-8 carbon atoms, and are typically at least about 12 atoms in length. The chains may be attached to the mineral acid head group through ester, ether, thioether, amide, or other stable linkages, as illustrated
5 variously in the Figure 1 compounds.

A second class of amphipathic compounds which have been examined herein are capable of solubilizing minoxidil in an aqueous medium, but fail to promote transdermal uptake of minoxidil. Several of the
10 compounds in this class are acidic vesicle-forming lipids, typically having charged phosphate or sulfate free acid head groups, a pK less than about 5, and diacyl, dialkyl or sterol lipophilic moieties. Exemplary compounds include the free acid forms of negatively
15 charged phospholipids, such as phosphatidylglycerol (PG), phosphatidylinositol (PI), and phosphatidic acid (PA), dialkyl phosphate compounds, such as dicetyl phosphate, and sterol mineral acids, such as cholesterol sulfate and taurocholic acid. Other phosphate, phosphonate,
20 sulfate, and sulfonate lipids containing two or more lipophilic chains or a sterol group through ester, ether, or amide linkages are also in this general class. Representative members of this class are illustrated in Figure 2. These are: taurocholic acid (2a), cholesterol
25 sulfate (2b), phosphatidic acid (2c), and phosphatidylglycerol (2d).

B. Preparing the Minoxidil Composition

According to an important property of the
30 amphipathic compounds, optimal solubilization of minoxidil by the amphipathic compound occurs at a pH of about 5 or less, where a significant fraction of the compound exists in free acid form. The solubility dependence of minoxidil on pH is illustrated in Figure

3, for a 20% by weight solution of laureth sulfo-succinate (Figure 2a compound). Between pH 7.0 and about 5.0, minoxidil solubility increases from about 1.5% to nearly 5%. Little improvement is seen as the pH
5 is lowered beyond about 4.5. For most purposes, a pH of about 5 is preferred, since good solubility is achieved, and skin irritation which may result from below-physiological pH is minimized.

In a preferred method for preparing the
10 composition, a portion of the amphipath is converted to a free acid form, and then "titrated" to the desired pH, e.g., pH 5.0, with the metal salt form of the compound. This approach is illustrated in Example 1, which describes the preparation of a 20 weight % laureth
15 sulfosuccinate solution having a final pH of 5. Here the disodium salt of the compound is converted to the free acid form by passage through a cation exchange resin. Mixing the free acid with the disodium salt, at a ratio of about 1:3, yields a pH 5.0 solution suitable
20 for solubilization of the minoxidil. It is appreciated that the free acid and salt components effectively buffer the solution at the selected pH, obviating the need for additional buffering components.

Alternatively, the compound may be converted
25 to or supplied in free acid form, then adjusted to the selected pH with a suitable base, such as NaOH. In another approach, the dispersion containing the amphipathic compound can be acidified, typically to between pH 1-2 before complexing with minoxidil. After
30 forming the desired complex, the dispersion can be titrated back to a suitable pH, typically between 4-6.

Optimal solubilization of the minoxidil in an aqueous formulation requires a molar concentration of amphipathic compound to minoxidil of at least about 1:1,

10.

and molar ratios of between 1:1 and 1:5 are typical. Figure 4 illustrates the increasing solubility of minoxidil with increasing concentration of amphipath in an aqueous solution at pH 5.0. Details are given in
5 Example 4. As seen from the figure, minoxidil solubility up to about 5 percent by weight was achieved at the highest amphipath concentration.

The minoxidil composition is preferably formed by adding dry minoxidil to the aqueous solution of
10 amphipath, prepared as above, to a desired pH and amphipath concentration. Typically, the solution is warmed to about 50°C, and the minoxidil is added slowly with stirring. When the minoxidil is completely dissolved, the solution is cooled and the pH adjusted,
15 if needed. The general method is illustrated in Examples 1 and 2, for the preparation of laureth sulfosuccinate/minoxidil compositions; in Example 5, for the preparation of a Crodafos™/minoxidil composition; and in Example 6, for the preparation of a taurocholic
20 acid/minoxidil composition. All of the compositions gave clear aqueous solutions.

As indicated above, the composition of the invention includes minoxidil in a substantially non-crystalline or molecular monodisperse or dissolved form.
25 These terms are defined herein to indicate that the minoxidil composition is substantially free of crystalline minoxidil, as judged, for example, by examination by polarization microscopy. It will be appreciated that the minoxidil may be present in a
30 microdispersion, such as in micellar or microemulsion form, and/or as a soluble molecular binary drug/amphipath complex. Thus, in preparing the composition, complete drug solubilization is judged by

the absence of the drug in crystalline or micro-crystalline form.

The composition may include additional soluble or suspension components, such as metal chelators, preservatives, and/or conventional lipid, emulsifying or gelling agents used in formulating ointment and cream topical formulations. Exemplary metal chelators include EDTA and DTPA, and exemplary preservatives include propyl- and methylparaben. Agents suitable for formulating the composition in cream or ointment form are known.

The use of the aqueous composition for topical administration of minoxidil, and transdermal uptake characteristics of compositions containing each class of amphipathic compound, are considered in Section IV below.

II. Minoxidil Composition in Lipophilic Solvents

According to another important aspect of the invention, the above-described amphipathic compounds which promote solubilization of the drug in aqueous medium, also promote solubilization in lipophilic solvents, such as chloroform, in which the drug is otherwise practically insoluble.

The amphipathic compound used in preparing the minoxidil composition in a lipophilic solvent may be supplied in free form, or converted to a free acid by treatment with a cation-exchange resin, as above. Typically, however, when the compound is supplied is the salt form, the compound is most conveniently converted to a free acid by solvent extraction into the organic solvent phase of an acidified, two-phase extraction mixture, such as in the Bleigh-Dyer extraction procedure detailed in Example 7. In this example, the amphipathic

compound is shaken in an acidified chloroform/methanol/water mixture and extracted in free acid form from the lower chloroform phase.

The solution of amphipathic compound
5 containing the free acid form of the compound in a lipophilic solvent (such as the lower-phase extract from a BleighDyer extraction) is mixed with dry minoxidil to form a non-crystalline dispersion of minoxidil in the solvent. For many amphipathic compounds, such as those
10 described in Examples 7-11, dispersion of the drug in a nonflocculated form also requires addition of a vesicle-forming lipid, such as PC, as illustrated in Examples 7-9, or an emulsion-forming agent, such as illustrated in Example 10 and 11. Vesicleor emulsionforming lipids
15 are also added to the solution when the composition is to be used in forming a liposomal or emulsion form of molecularly dispersed minoxidil, as detailed in Section III below. Solubilization of minoxidil in the lipophilic solvent containing an amphipathic compound
20 may also be achieved by addition of a co-solvent, such as an alcohol or glycol, to the mixture of minoxidil and amphipath in lipophilic solvent.

One application of the minoxidil/lipophilic solvent mixtures is for delivery of the drug in spray
25 form from a self-propelled atomizer system. Here minoxidil, the amphipathic compound and, if needed, a vesicle- or emulsion-forming agent are co-dissolved in a Freon chlorofluorocarbon solvent. Several fluorochloro-carbon propellant solvents have been used or proposed
30 for self-contained spray devices. Representative solvents includes "Freon 11" (CCl_3F), "Freon 12" (CCl_2F_2), "Freon 22" (CHClF_2), "Freon 113" ($\text{CCl}_2\text{FCClF}_2$), "Freon 114" ($\text{CClF}_2\text{CClF}_2$), and "Freon 115" (CClF_2CF_3), as

well as other fluorochloro substituted methyl and ethyl compounds.

The propellant solution is loaded in a conventional pressurized propellant spray device for delivering a metered amount of spray-dried minoxidil dispersed in the propellant. Since the spray system may require long-term storage of the solution components in the propellant solvent, the lipid components in the system should be selected for stability on storage, for example by employing partially or totally saturated amphipathic and vesicle-forming lipid components.

In use, the propellant spray device produces a fine-particle spray of solubilized minoxidil which is directed against the skin area being treated. The spray particles initially contain minoxidil complexed with the amphipathic compound and, if present, added co-solubilizing agent, dispersed in the propellant solvent. Rapid evaporation of the solvent, as the spray particles are ejected through the air, yields noncrystalline minoxidil particles which form a layer of drug particles which substantially cover the sprayed skin area.

III. Minoxidil Composition in Lipid Bodies

According to another aspect of the invention, the minoxidil composition can be entrapped in non-crystalline form in both lipid emulsion particles and liposomes, providing additional advantages for topical administration of the drug. One of these advantages, in the case of liposomes, is the ability to modulate the rate of drug release from the composition, by selection of the suitable vesicle-forming lipids. Another is the greater drug loading capacity of lipid particles. Liposomes and emulsion particles are both compatible with topical ointment and cream formulations, and in

fact are commonly added to skin creams as moisturizing agents. Liposomes may also be adapted for use with self-propelled spray systems, providing a convenient method of delivery of a non-crystalline, high-
5 concentration minoxidil composition.

Considering first the preparation of non-crystalline minoxidil liposomes, the vesicle forming lipids are preferably neutral phospholipids, such as PC, and may also include negatively charged phospholipids,
10 such as PG, phosphatidylinositol (PI), and phosphatidylserine (PS) which can function as the negatively charged amphipaths in the composition. For the reasons discussed above, however, the amphipathic compound used in the composition is preferably a single chain mineral
15 acid compound of the type which by itself would not form lipid bilayer vesicles upon hydration. Other liposomal lipids, such as cholesterol, may also be included.

Studies conducted in support of the present
20 invention indicate that minoxidil transdermal uptake can be modulated by the factors which affect the fluidity of liposome membranes, such as the extent of phospholipid acyl chain saturation. As a rule, transdermal uptake is decreased by entrapment of the non-crystalline
25 composition in liposomes. Another factor which is important in rate of drug uptake is the ratio of vesicleforming lipids to minoxidil. Preferred weight ratios of phospholipid to minoxidil are between about 1:1, which gives relatively high transdermal uptake and
30 4:1, which gives quite low uptake.

The non-crystalline liposome composition can be formed by a variety of methods which are modifications of existing liposome preparation methods. For example, to prepare the liposomes by lipid

-15-

hydration, a lipid solution containing minoxidil, the amphipathic compound, and PC, is prepared as above, by dissolving the lipid and minoxidil in the lower-phase solution of amphipath in free acid form. The resulting solution of minoxidil, amphipath, and lipid are dried to a thin film, then hydrated with a suitable aqueous buffer. This hydration method is illustrated in Examples 7-9 below. Alternatively, a film of vesicle-forming lipids alone can be hydrated by a solution of the non-crystalline minoxidil composition, formed as in Section I.

One preferred method of forming the liposome composition uses a novel lipid injection method described in co-owned patent applications for "High-Encapsulation Liposome Processing Method", Serial No. 908,765, filed September 18, 1986, and "High-Concentration Liposome Processing Method", Serial No. 909,122, filed September 18, 1986. In this method, a solution of minoxidil, amphipathic compound, and vesicleforming lipids in a preferably chlorofluorocarbon solvent is prepared as described above in Section II. This solution is injected into an aqueous medium under selected temperature and pressure conditions which lead to liposome formation. According to an important feature of the method, solvent injection may be continued, with or without concomitant liposome sizing, until a liposome composition having the consistency of a thick paste is formed. The paste composition has the capability of high minoxidil loading, and also is suitable as a cream or ointment without further processing.

The liposome composition may also be delivered in dried particle form from a self-propelled spray device. Here the liposomes, formed according to above

5 methods, are dried, by spray drying, then suspended in a
chlorofluorocarbon propellant solvent. Methods for
spray drying liposomes and forming stable liposome-
particle suspensions in several Freon propellants, have
10 been described in co-owned U.S. patent application for
"Liposomes for Inhalation", filed February 2, 1987, and
similar methods are applicable to the liposomal
composition of the present invention. Studies conducted
in support of the just-cited patent applications have
15 examined the stability and size characteristics of spray
dried liposomes in several Freon propellants. Good
liposome stability, as measured by microscopic
examination of the liposomes and retention of
encapsulated material was seen with Freons 12, 113, 114,
and 115.

The suspension of dried minoxidil liposomes in
propellant solvent can be administered in metered dose
spray form from a conventional pressurized spray device
such as used above for delivery of a Freon dispersion
20 solution of minoxidil/amphipath components.

Methods for producing a non-crystalline lipid
emulsion composition, according to the invention, may
similarly follow standard preparative methods, with
modification to include the amphipathic compound needed
25 for minoxidil solubilization. Examples 10A and 10B
illustrate methods for forming emulsions of lysoPA/PA
and PA alone in a Tween-20™ emulsion, respectively.
Briefly, a Bleigh-Dyer solvent extraction containing the
free acid form of the lysoPA/PA mixture or PA alone was
30 added to minoxidil, and the lipid solution dried to a
thin film. Hydration of the lipid film with an aqueous
buffer containing 30% Tween-20™ yielded an aqueous non-
crystalline emulsion.

-17-

The minoxidil composition of the present invention may also be formulated as a stable microemulsion, i.e., a clear, thermodynamically stable emulsion. Typically, the microemulsion is formed by first producing a water-in-oil microemulsion composed of conventional surfactant and oil-phase components and the amphipathic compound needed for minoxidil solubilization. This is done conventionally, e.g., by combining the oil-phase, surfactant and amphipath components at a suitable temperature, e.g., 60°C, with stirring. After the mixture clears, it is acidified (if it is not otherwise in acid form), and minoxidil is added slowly with stirring until the mixture becomes clear again. The final clear microemulsion can be adjusted to pH 5.0 without destabilizing the dispersion. Example 11A illustrates a 4% water-in-oil microemulsion formed according to this general procedure.

An oil-in-water microemulsion can be formed readily by adding water dropwise to the above microemulsion, until a desired minoxidil concentration is reached. This procedure is illustrated in Example 11B below, which describes preparation of a 2% minoxidil oil-in-water microemulsion. An oil-in-water emulsion may also be prepared directly by increasing the concentration of water included in the microemulsion formed before addition of minoxidil.

Alternatively, the emulsion may be formulated in a concentrated, viscous or paste-like form for topical administration, giving the same advantages of high loading available with the liposomal formulation. Alternatively, the emulsion composition can be produced in a self-propelled device by dissolving the minoxidil/amphipath/emulsion lipid components in a Freon solvent, as above, and delivering the components in

spray form. Non-crystalline minoxidil/lipid particles are formed during rapid evaporation of the propellant solvent.

5 IV. Utility

A. Transdermal Uptake Characteristics

The transdermal uptake characteristics of several exemplary minoxidil compositions prepared according to the invention have been examined, as outlined generally in Example 12. Briefly, a small isolated skin patch is sealed between upper and lower chambers of a transdermal cell, and an aliquot of the selected composition (spiked with radiolabeled minoxidil) is applied to the upper surface of the patch. The lower chamber holds a reservoir buffer which is in contact with the lower surface of the skin, and which is circulated through the lower chamber by a constant-rate pump. As drug penetrates the skin patch, it is captured in the lower reservoir, and pumped out of the chamber into assay vials for scintillation counting. Transdermal uptake (drug penetration) is typically measured for over a 24 hour period.

The preparation of the several compositions studies is detailed in Examples 1, 2, and 5-11. The control drug composition used in the studies is a 2% minoxidil composition in an ethanol/propylene glycol/water vehicle. The transdermal uptake of this control formulation, over a 24 hour period, is shown by the open squares in Figure 5. The cumulative amount of drug taken across the skin in the 24 hour period is less than about 1% of the total applied to the skin.

Also shown in the figure are the transdermal uptake curves for a soluble 2% minoxidil/laureth

-19-

sulfosuccinate composition (crosses), and a soluble 5% minoxidil laureth sulfosuccinate composition (open diamonds). As seen, both of the soluble laureth sulfosuccinate compositions gave higher transdermal penetration rates than the control composition, expressed as μg drug penetrating/ cm^2 of skin. The total cumulative drug uptake for the 2% composition is about 6% of the total applied to the skin.

Interestingly, the 2% laureth sulfosuccinate composition, which contains about 8% by weight of the laureth sulfosuccinate, gave a significantly higher transdermal uptake than the 5% composition, which contains about 25% by weight of the amphipath. This result may be due to the greater viscosity of the 5% composition, and/or interactions between the surfactant and skin which are less inhibitory in the 2% composition.

Figure 5 also shows the transdermal uptake for a soluble 2% minoxidil/taurocholic acid composition (open triangles). It is evident that taurocholic acid does not promote the uptake of minoxidil across the skin, even though it is effective in solubilizing the drug in an aqueous formulation.

Figure 6 shows similar transdermal uptake data comparing a minoxidil/crofadol composition prepared according to the invention with the above control composition. The transdermal uptake for the composition of about $700 \mu\text{g}/\text{cm}^2$ is nearly three times that of the 2% laureth sulfosuccinate composition and about 35 times that of the control material. The total amount of drug material passing through the skin in a 24 hour period is about 35% of the total added.

The transdermal uptake characteristics of several liposomal and emulsion compositions were also

-20-

examined, and compared with the above minoxidil control composition. Figure 7 shows transdermal uptake curves for 2% (crosses) and 5% (open diamonds) minoxidil/lauryl sulfosuccinate/PC liposome suspension formed as in Example 7. The data show enhanced transdermal uptake when compared with the control formulation (open squares). It is noted that, in contrast to the results observed for the soluble laureth sulfate composition, the higher percent composition (5% minoxidil) gave greater transdermal uptake. Both liposome formulations gave about 5% total drug penetration over the 24 hour test period. It is also noted that the best liposomal formulation gave higher drug penetration (about 350 $\mu\text{g drug/cm}^2$) than the best laureth sulfosuccinate composition (about 240 $\mu\text{g drug/cm}^2$).

The figure also shows transdermal uptake for a 1% minoxidil/cholesterol sulfate/PC liposome composition. This composition thus differs from the ones just discussed in that cholesterol sulfate has been substituted for lauryl sulfosuccinate. As seen, virtually no transdermal uptake of minoxidil occurred during the 24 hour test period.

In Figure 8, the transdermal uptake of the above 2% minoxidil/lauryl sulfosuccinate/PC liposomes (crosses) is compared with that of 2% minoxidil/oleamido PEG-2 sulfosuccinate/PC liposomes (open diamonds). As observed, the latter composition gave substantially higher transdermal uptake than either 2% or 5% minoxidil/lauryl sulfosuccinate/PC liposome compositions.

Finally in Figure 9 are shown transdermal uptake curves for suspensions formed from 2% minoxidil and either lysoPA/PA (crosses) or PA alone (open diamonds), according to the preparative methods of

-21-

Examples 10 and 11, respectively. The control composition is indicated by open squares. PA alone gives very poor drug uptake, whereas with the addition of lysoPA, uptake is enhanced slightly above the control level.

Considering the data as a whole, it is seen that enhanced transdermal uptake is present in each composition where the amphipathic compound used to solubilize minoxidil contains a single lipophilic chain and a sulfosuccinate (sulfate), phosphonate, or phosphate polar head. In all cases where the lipophilic moiety of the amphipath was either a sterol or included more than a single lipophilic chain, transdermal uptake was severely limited.

B. Drug Administration

The composition of the invention may be administered topically in a water-base cream, ointment, or gel form. Several factors contribute to cosmetic advantages of the water-base composition. High drug loading up to 5% or more in non-crystalline form is possible, and the drug remains in a non-crystalline form up to several hours or more after administration, since solvent evaporation is relatively slow and because evaporation can occur without leading to drug crystallization. Since the composition contains little or no alcohol, it can be applied without stinging.

The drug is efficiently taken up by the skin, by virtue of the selected amphipath, and different rates of uptake can be achieved by varying the drug or amphipath concentration, and by selection of different amphipathic compounds.

The water-base composition may additionally contain liposome or lipid-emulsion particles in which

the drug can be entrapped in non-crystalline form. In one embodiment, a concentrated liposomal composition having desired a desired cream or ointment consistency can be formulated using a novel solvent injection system. The liposome formulation can have high loading, and rate of drug uptake can be modulated by choice of lipid components and relative molar amounts of lipid and drug. The lipid formulation is also expected to have the known moisturizing benefits of topical lipid formulations.

In another embodiment, the composition is dispersed in a chlorofluorocarbon solvent for delivery in spray form. The spray form has many advantages of the water-base formulation, including high drug loading and enhanced drug uptake. Additionally, the spray composition has the advantage that it can be applied in a more convenient manner and without matting the hair in the treated scalp region.

The following example illustrate methods of preparing non-crystalline minoxidil compositions according to various embodiments of the invention, and compares transdermal penetration characteristics of the various compositions. The example are intended to illustrate, but not limit the scope of, the invention.

Materials

Disodium laureth sulfosuccinate was obtained from Sherex (Dublin, CA), and supplied under the trade name "Varsulf SBFA-30" or from Mona Industries (Patterson, NJ), and supplied under the trade name "LEO-40"; disodium lauryl sulfosuccinate, from Mona Industries; disodium salt of taurocholic acid, from Sigma Chemical Co. (St. Louis, MO); oleamido-

polyethylene glycol-2 sulfosuccinate, disodium salt, from Mona Industries; Crodafos™ N3 acid (oleth 3 phosphate), from Croda, Inc. (Fullerton, CA); phosphatidic acid (PA), from Avanti Polar Lipids, Inc. (Birmingham, AL); Tween-0 from J.T. Baker (Phillipsburg, NJ); and cholesterol sulfate, from Sigma Chemical Co. (St. Louis, MO). Minoxidil, USP, was obtained from Upjohn (Kalamazoo, MI); methylparaben and propylparaben, from Sigma Chemical Co (St. Louis, MO); diethylenetriaminepentaacetic acid (DTPA), from Aldrich (Milwaukee, WI); and partially hydrogenated egg phosphatidylcholine (PC), from Asahi (Tokyo, Japan). MES (morpholinoethane sulfonic acid) was obtained from Sigma (St. Louis, MO). AG 50W-X8 cation exchange resin were supplied by Bio-Rad (Rockville Center, NY). Polyethoxylated (C8-C10) glycerides (Labrosol™), stearyl alcohol ester of stearic acid (isostearate D'isostearique), and polyglycerol isostearate (plurol isostearique) were obtained from Gattefosse™ (Elmsford, NY).

Example 1

5% Minoxidil/Laureth Sulfosuccinate Composition

A. Preparing the free acid (laureth sulfosuccinic acid) from disodium laureth sulfosuccinate.

250 ml distilled water was added to 750 ml of a 40% (wt/vol) solution of disodium laureth sulfosuccinate to make one liter of 30% solution. This solution was passed over a prepared column packed with approximately 270 g of AG50W-X8 cation exchange resin that has been converted to the hydrogen ion form. The eluate was collected as a single fraction after the pH

-24-

of the eluate dropped to 1.3. This eluate was the free acid, lauryl sulfosuccinic acid.

5 B. Complexing minoxidil with the hemiester sulfosuccinate.

800 ml of the AG50W-X8 eluate from above was mixed with 2,400 ml of a 30% (wt/vol) solution of disodium laureth sulfosuccinate. The resulting solution was heated to about 50°C and 192 grams minoxidil was
10 added slowly with stirring. After complete dissolution of the minoxidil occurred, the solution was cooled to room temperature, and the pH adjusted to 5.0 ± 0.1 . Distilled water sufficient to bring the volume to 3,840 ml was added, yielding a clear, 5% minoxidil/25%
15 surfactant solution which is free of microcrystals, as judged in the polarizing microscope (630x magnification).

The pH of the final dispersion can be varied by changing the ratio of laureth sulfosuccinic acid to
20 disodium laureth sulfosuccinate. A 1:3 ratio (as above) after minoxidil dissolution gave a pH of about 4.7 ± 0.2 .

Example 2

25

2% Minoxidil/Laureth Sulfosuccinate Composition

A 2% dispersion of solubilized minoxidil and the free acid of laureth sulfosuccinate was prepared substantially as in Example 1, with the following
30 modifications: The AG50W-X8 column was prepared with 100 g. Two hundred fifty ml of 40% wt/vol disodium laureth sulfosuccinate was diluted to 20% wt/vol surfactant by adding 250 ml distilled water. This solution was passed over the AG50W-X8 cation exchange

-25-

column and the free acid eluate collector. Four hundred ml of free acid solution was combined with 1,200 ml of 20% disodium laureth sulfosuccinate and 2.0 liters distilled water. The mixture was heated to 50°C and 80 grams minoxidil was added slowly with mixing. After minoxidil dissolution, other excipients may be added. The mixture was cooled to room temperature, and the pH adjusted to about 5.3 ± 0.1 . Distilled water was added to give 4.0 l of a clear dispersion containing 2% solubilized drug and 8% laureth sulfosuccinate.

Example 3

Minoxidil Solubility: pH Dependence

A 20% solution of the free acid of laureth sulfosuccinate in distilled water was prepared as described in Example 1A. More acidic solutions of laureth sulfosuccinate were prepared by increasing the proportion of free acid in the free acid/disodium salt mixture, and more basic forms, by decreasing the ratio. The different-pH solutions were each heated to about 50°C and dry minoxidil containing tritiated minoxidil was added slowly with stirring until minoxidil saturation was achieved. The dispersions were cooled overnight at 4°C and centrifuged. The concentration of minoxidil in the clear solution was determined by scintillation counting. The results, expressed in mg minoxidil/ml laureth sulfosuccinate solution, are plotted in Figure 3 for two separate experiments. As seen, minoxidil solubility is very low at pH 7.0, and increases linearly to a maximum at a pH about 4.5-5.0.

-26-

Example 4Minoxidil Solubility:Dependence on Amphipath Concentration

5 Solutions of the free acid of laureth
sulfosuccinate, at concentrations of 0, 5%, 10%, 15%,
20%, and 25% by weight in distilled water were prepared
as in Example 1A. Each solution was heated to about
10 50°C, and radiolabeled minoxidil was added slowly with
stirring until minoxidil saturation was achieved, this
being monitored as described in Example 3. The pH of
each solution was adjusted to about pH 5 prior to
centrifugation and scintillation counting. The results,
expressed in mg minoxidil/ml laureth sulfosuccinate
15 solution, are plotted in Figure 4. Minoxidil solubility
in the absence of the amphipath is about 3 mg/ml, or
0.3%. With increasing concentrations of the laureth
sulfosuccinate, up to 25 weight percent, the solubility
if minoxidil increases up to about 50 mg/ml, or 5% at
20 pH 5.

Example 5Minoxidil/Crodafos™ Composition

25 Five milliliters of Crodafos™ N3 acid (the
oleth-3 phosphate) was obtained in free acid form and
diluted in 5 ml distilled water: 10 ml punctilious
ethanol. The solution was heated to 37°C, 2 g minoxidil
was added slowly with stirring until dissolution. The
30 mixture was diluted with 180 ml of aqueous solution
buffered by MES at pH 5.5. Examination of the
composition with a polarizing microscope showed no drug
crystals in the dispersion containing 1% minoxidil and
5% Crodafos™ N3 acid.

Example 6Minoxidil/Taurocholic Acid Composition

5 Taurocholic acid, sodium salt, was converted
to a free acid form by the procedure of Example 11.
About 100 mg by weight of the free acid form was mixed
with 20 mg of dry minoxidil in 1 ml pH 5.5 buffer,
yielding a clear solution with a final minoxidil
10 concentration of about 2% by weight. Stirring was
continued until a clear solution was obtained, after
which the pH of the solution was readjusted to 5.0. No
crystals were observed on examination of the composition
with a polarizing microscope.

15

Example 7Minoxidil/Lauryl Sulfosuccinate/Liposome Composition

20 This example describes the preparation of a
minoxidil/lauryl sulfosuccinate/liposome composition.
The free acid form of disodium lauryl sulfosuccinate was
formed by the Bleigh-Dyer extraction procedure, as
follows: A first solvent mixture was prepared by mixing
8 ml 1 N HCl, 20 ml methanol, and 10 ml chloroform in a
250 ml separatory funnel. To this solvent was added 400
25 mgs of the disodium lauryl sulfosuccinate, which was
dissolved by vigorous shaking. A second solvent mixture
containing 7 ml HCl, 10 ml chloroform, and 3 ml
distilled water was added to the funnel, which was then
30 shaken vigorously, and allowed to phase separate.

The lower chloroform phase (containing the
bulk of the free acid form of the lauryl sulfosuccinate)
was collected in a 250 ml round bottom flask containing
580 mgs partially hydrogenated PC (PHPC) and 200 mgs of

-28-

minoxidil, both in dry form. The flask was swirled until both of the dry components were in solution, and to this solution was added butylated hydroxy toluene (BHT) in chloroform to a final concentration of about 1
5 mM.

The upper phase in the separatory funnel was reextracted with 5 ml chloroform and 1 ml methanol by vigorous shaking, and the lower phase which formed on standing was collected in the flask containing the
10 minoxidil/lipid solution. The solvent in the flask was removed by rotary evaporation, yielding a thin lipid film. The dried material was further lyophilized for 1/2 hr to insure complete solvent removal.

The final minoxidil/liposome suspension was
15 formed by hydrating the lipid film with 10 ml of MES buffer, pH 5.5, containing 0.01 % DTPA, using a mechanical "wrist" shaker. Shaking for 1 hour with the flask maintained at 50° C over a water bath was sufficient to produce complete hydration, as judged by
20 the uniform appearance of the liposome suspension. The pH of the suspension, which had dropped to about 4 during the hydration step, was raised to 5.0 with 5 N NaOH. Microscopic examination of the liposome
25 suspension showed a heterogeneous-size population of spherical vesicles. No polarizing crystals were noted.

Example 8

Minoxidil/Oleamido Sulfosuccinate/Liposome Composition

30 Oleamido-(2 polyethylene glycol)-sulfo-succinate was converted to the free acid form by Bleigh-Dyer extraction procedure, substantially as described in Example 7. The first lower chloroform phase was collected in a 250 ml round bottom flask

-29-

containing 580 mgs partially hydrated PC (PHPC) and 200 mgs of minoxidil, both in dry form. To this was added the second lower extraction phase, as above, and the amphipath/minoxidil/lipid solution was taken to dryness with rotary evaporation and lyophilization.

The final minoxidil/liposome suspension was formed by hydrating the lipid film with 10 ml of MES buffer, pH 5.5, containing 0.01 % DTPA, under hydration conditions used in Example 7. The pH was adjusted to 5.0 after hydration was completed. Microscopic examination of the liposome suspension showed a heterogeneous-size population of spherical vesicles. No polarizing crystals were noted.

15

Example 9

Minoxidil/Cholesterol Sulfate/Liposome Composition

Cholesterol sulfate was converted to the free acid form by cation exchange chromatography in a methanol/chloroform/water (5:4:1) solvent. The free acid form (120 mgs) was dissolved in 2 ml of chloroform, and this solutions was added to a round bottom flask containing 40 mg minoxidil and 370 mgs of PC. The flask was gently agitated until the components were completely dissolved. The solution was dried to a thin lipid film in a round bottom flask with rotary evaporation and lyophilization, as above. A liposome suspension was prepared as in Example 8. No minoxidil crystal were observed in the suspension.

30

Example 10A. Minoxidil/LipoPA-PA Composition

5 A mixture of lipophosphatidic acid (lipoPA)
and PA was formed by long term storage of pure PA at
4°C. The mixture was confirmed with thin layer
chromatography as containing significant portions of
both PA and lipoPA (Figure 2i). One hundred ten mg of
the mixture was converted to the free acid form by the
10 Bleigh-Dyer extraction procedure above, and the combined
lower-phase extracts were added to a round bottom flask
containing 30 mg minoxidil. The flask was gently
agitated until the drug was completely dissolved. The
solution was dried to a thin lipid film in a round
15 bottom flask with rotary evaporation and lyophilization,
as above.

The final minoxidil/lysoPA/PA liposome
suspension was formed by hydrating the lipid film with
10 ml of MES buffer, pH 5.5, also containing 30%
20 Tween-0, under hydration conditions used in Example 7.
The pH was adjusted to 5.0 after hydration was
completed. Microscope examination of the suspension
showed a very small (< 1 micron diameter) particles. No
polarizing crystals were noted.

25

B. Minoxidil/PA Composition

Freshly obtained PA was examined by thin layer
chromatography for purity, and only minor contaminants
were observed. One hundred ten milligrams PA was
30 converted to the free acid form by the Bleigh-Dyer
extraction procedure above, and the combined lower-phase
extracts were added to a round bottom flask containing
30 mg minoxidil. An suspension was formed by hydration
of the dried-film lipids, as in Example 9. No minoxidil

crystals were observed when examined with polarization microscopy.

Example 11

5

Minoxidil Microemulsions

A. Water-in-oil Microemulsion

Labrasol™ (6.6 g), isostearate D;isostearique (0.8 g), plurol isostearique (2.8 g) (all obtained from Gattefosse'), and disodium laureth sulfosuccinate (4.0 g) and 2.03 g distilled water were mixed together in a 50 ml glass beaker, and the mixture stirred in a 54°C water bath until clear. The pH of the mixture was lowered to between 1-2 with a 1 N HCl. Minoxidil (0.8 g) was slowly added to the mixture at a temperature of about 56°C. Addition was complete after 10 minutes. After an additional 10 minutes of incubation, at a final temperature of about 60°C, the suspension was clear. The pH was then raised to about 5.0 with 5 N NaOH. Incubation with stirring was continued for another 15 minutes, to a final temperature of about 64°C. The material was then allowed to cool to room temperature, and a final adjustment to pH 5 was made. The final dispersion was a clear, yellow oily microemulsion containing 4% (by weight) minoxidil. The maximum amount of minoxidil which can be added, consistent with a clear microemulsion, is between about 5-7% by weight.

B. Oil-in-water Microemulsion

30 Distilled water was added dropwise to 10.57 g of the 4% minoxidil water-in-oil emulsion from part A above. A total of about 12 g of DW was added, to a final minoxidil concentration of about 2%. As DE was added, the clear oily suspension became moderately

viscous, and finally, a clear liquid suspension which gave a cooling effect on the skin (diagnostic indication for oil-in-water microemulsion). The clear oil-in-water emulsion was stable on storage.

5

Example 12

Transdermal Uptake Studies

10

A. Experimental protocol

The transdermal cell used for measuring skin penetration has upper and lower chambers which are separated by a skin patch. The lower chamber is designed to permit continuous flow through of saline, which collects drug penetrating from the outer side of the skin (exposed to the upper chamber) through the skin and into the saline in the lower chamber. An infusion pump is used to move through the chamber at a controlled rate (about 5 ml/hour).

20

Female hairless mice, strain HRS/hr, were obtained from Simonsen (Gilroy, CA). The animals were 7-8 weeks old, and weighed 20-30 gm when used. After sacrifice, three 2 cm diameter skin patches were removed from each animal. The patches were individually mounted in the cell, and held sealed against the lower chamber by an O-ring which is pressed against the patch by clamping.

25

Prior to adding the drug solution to the skin, a phosphate buffered saline solution was pumped through the system, at a flow rate of about 5 ml/hr for one hour. Fractions were collected continuously from the outlet side of the lower chamber, and dispensed into vials in a fraction collector. Collection time per fraction was one hour. Fractions were collected for up

30

-33-

to 24 hours after the drug solution was applied to the skin membrane. After the test period, the skin patch is washed several times, and removed. The hourly fractions, wash fractions obtained at the end of the experiment, and the skin patch itself were counted for radioactivity by conventional scintillation counting methods.

B. Control skin penetration test

The control vehicle was Rogaine™, obtained from Upjohn Co. This formulation contains 2% minoxidil in an ethanol/propylene glycol/water solvent vehicle, and was labeled with tritiated minoxidil before testing. One hundred fifty μ l samples were applied to skin patches and the uptake of minoxidil across the skin monitored as described. Typical results for a 24 hour test period are shown in Figure 5, where the control drug data is indicated by the open squares in the figure. As seen, the rate of uptake of the drug in the control formulation is substantially linear over the test period, and reaches a cumulative maximum, at the end of the test period, of about $30 \mu\text{g}/\text{cm}^2$, corresponding to about 0.5-1.0% of the total drug applied to the skin.

25

Example 13

Transdermal Penetration:

Laureth Sulfosuccinate Compositions

The 5% and 2% minoxidil/lauryl sulfosuccinate compositions prepared as in Examples 1 and 2, respectively, were tested for transdermal uptake, using the experimental methods described in Example 10. Three duplicate runs were made with each of the two

30

-34-

formulations, along with the control formulation (Example 12). The results, expressed in terms of cumulative μg drug uptake/ cm^2 of skin patch, are shown in Figure 5, where the data for the 2% composition is indicated by crosses, and for the 5% composition, by open diamonds. The open squares indicate uptake data for the control system, as indicated above.

It is seen that both sulfosuccinate compositions give greater drug transdermal penetration than the control drug formulation. The final cumulative doses correspond to about 0.5-1.0 for control drug, 2% for the 5% composition and 5-6% for the 2% composition.

Example 14

Transdermal Uptake: Crodafos Composition

A 1% minoxidil/Crodafos™ N3 acid was prepared as in Example 5, and tested for transdermal uptake using the experimental methods described in Example 12. These results are from four replicate cells, and are plotted along with the control formulation (Example 12). The results, expressed in μg drug penetration/ cm^2 skin, are shown in Figure 6, where the data for the 1% minoxidil in Crodafos™ N3 acid is indicated by crosses; open squares indicate the control formulation.

It is seen that the Crodafos™ formulation causes more than an order of magnitude increase in cumulative drug uptake as compared to the control formulation, even though the control formulation has twice the drug loading.

-35-

Example 15Transdermal Uptake: Taurocholic Acid Composition

5 The 2% minoxidil/taurocholic acid compositions prepared as in Example 6 were tested for transdermal uptake using the experimental method described in Example 12. Four replicate cells were run and the data is plotted in Figure 5 as open triangles. The open squares indicate transdermal penetration by the control
10 formulation. It can be observed that although taurocholic acid has the same drug loading as the control formulation, less drug is put through the skin.

Example 16

15

Transdermal Uptake:Lauryl Sulfosuccinate Liposome Composition

The 2% and 5% minoxidil/lauryl sulfosuccinate liposome compositions prepared as in Example 7 were
20 similarly tested for transdermal delivery, with the results therein in Figure 7. The control formulation is denoted by open squares, the 2% liposome formulation by crosses, and the 5% liposome formulation by open diamonds. The 2% formulation delivers 6 times the
25 cumulative dose of the control formulation at 24 hours, and the 5% formulation delivers 13 times the cumulative dose of the control at 24 hours.

Example 17

30

Transdermal Uptake:Oleamido PEG-2 Sulfosuccinate Liposomes

A 2% minoxidil/oleamido PEG-2 sulfosuccinate liposome composition prepared as in Example 8 was tested

-36-

for transdermal delivery of minoxidil using the experimental protocol described in Example 12. The data are plotted in Figure 8 as open diamonds, and compared to the control formulation (open squares) and the 2% minoxidil lauryl sulfosuccinate liposomes (crosses).

The oleamido PEG-2 composition produces an approximately threefold increase in transdermal uptake over the lauryl sulfosuccinate liposome preparation. It is evident that changing the hemiester sulfosuccinate in the composition can change the rate of transdermal delivery of drug.

Example 18

15

Transdermal Uptake:

Cholesterol Sulfate Liposome Composition

The minoxidil/cholesterol sulfate/PC composition was prepared as in example 9 and assayed for transdermal drug delivery as in Example 12. The results are plotted as open triangles in Figure 8. As seen, this composition showed no transdermal penetration, even though the composition was saturated with drug (1% wt/vol).

25

Example 19

Transdermal Uptake: LysoPA/PA Liposome Composition

The minoxidil/lysoPA/PA in 30% Tween-0™ composition prepared as in Example 11 was tested for transdermal drug delivery as in Example 12. The data plotted as crosses in Figure 9. This composition is similar to the control formulation during the first 12 hours, and then increases substantially relative to the control formulation during the next twelve hour period.

Example 20Transdermal Uptake: PA Liposome Composition

5 The minoxidil/PA in Tween-0 composition was
prepared as in Example 11, and assayed for transdermal
drug penetration as in Example 12. These data are also
plotted as open diamonds in Figure 10. This composition
delivers one-third the drug of the control formulation
10 (open squares) and one-sixth that of the composition
containing lyso PA (crosses). The results indicate that
lyso PA and not PA or Tween-0™ is responsible for
facilitating transdermal delivery of the drug.

15 Although the invention has been described with
reference to particular methods of preparation, modes of
drug administration, and transdermal uptake
characteristics, it will be appreciated that various
modifications and changes in the methods and results can
20 be made or achieved within the scope of the invention.

25

30

-38-

IT IS CLAIMED:

1. A non-crystalline minoxidil composition comprising minoxidil complexed with an amphipathic compound having a pK less than about 5 and containing a single lipophilic chain moiety and a polar head group moiety selected from a sulfate, sulfonate, phosphate and phosphonate group.
2. The composition of claim 1, wherein the molar ratio of amphipathic compound to minoxidil is at least about 1:1, and the pH is between about 4-6.
3. The composition of claim 1, wherein the amphipathic compound ester is selected from the group consisting of sulfosuccinic acid hemiesters, alkyl phosphonates, and alkyl phosphate esters.
4. The composition of claim 3, wherein the organic acid monoalkyl ester is an ethoxylated sulfosuccinic acid hemiester.
5. The composition of claim 1, which further includes phospholipids at a phospholipid to minoxidil molar ratio of between about 1:1 and 4:1.
6. The composition of claim 1, wherein the minoxidil and amphipathic compound are dispersed in non-crystalline form in a chlorofluorocarbon solvent.
7. The composition of claim 6, for use in forming a suspension of liposomes containing minoxidil in substantially monomolecular form, which further includes a phospholipid, at a phospholipid to minoxidil

molar ratio of between about 1:1 and 4:1, wherein the composition is injected into an aqueous medium under conditions which result in liposome formation in the aqueous medium.

5

8. The composition of claim 1, which further includes oil-phase and surfactant components and the composition is a microemulsion.

10

9. The composition of claim 1, for use in administering minoxidil topically in ointment form, which further includes at least about 60% by weight of water, and minoxidil is present at a weight concentration of between about 1-5%.

15

10. The composition of claim 9, which further includes phospholipids, at a phospholipid to minoxidil molar ratio of between about 1:1 and 4:1.

20

11. The composition of claim 9, which further includes oil-phase and surfactant components and the minoxidil is present in microemulsion form.

25

12. A method of applying minoxidil topically in a non-crystalline form which remains non-crystalline at least several hours after application to skin, comprising complexing the minoxidil with an amphipathic compound having a pK less than about 5 and containing a single lipophilic chain moiety and a polar head group moiety selected from a sulfate, sulfonate, phosphate and phosphonate group.

30

13. The method of claim 12, wherein the amphipathic compound is selected from the group

-40-

consisting of sulfosuccinic acid hemiesters, alkyl phosphonates, and alkyl phosphate esters.

5 14. The method of claim 12, for use in
applying minoxidil in ointment form, which further
includes formulating the minoxidil and amphipathic
compound in an aqueous medium, to a final minoxidil
weight concentration of between 1-5%, and a final weight
concentration of water of at least about 60%

10

 15. The method of claim 12, for use in
administering minoxidil in spray form, which further
includes dispersing the minoxidil and amphipathic
compound in a fluorochlorocarbon solvent, and spraying
15 the resultant mixture from valved cannister containing
the mixture under pressure.

 16. The method of claim 12, which further
includes adding a phospholipid to the mixture, at a
20 phospholipid to minoxidil molar ratio of between about
1:1 and 4:1, to form a liposomal minoxidil composition.

 17. The method of claim 12, wherein said
complexing includes first forming a stable water-in-oil
25 microemulsion containing oil-phase, surfactant and
amphipathic compound components, then adding minoxidil
to the microemulsion to a final minoxidil concentration
of between about 2-7 percent by weight.

30 18. The method of claim 17, which further
includes, after addition of minoxidil to the
water-in-oil microemulsion, adding water until an
oil-in-water microemulsion forms.

19. A non-crystalline minoxidil composition in which minoxidil is:

(a) present at a weight concentration of between about 1-5%;

5 (b) complexed with an amphipathic compound having a pK less than about 5, at an amphipath:minoxidil molar ratio of at least about 1:1;

(c) dispersed in substantially non-crystalline form in an aqueous or lipophilic solvent; and

10 (d) maintained in non-crystalline form for a period of at least several hours after application to the skin.

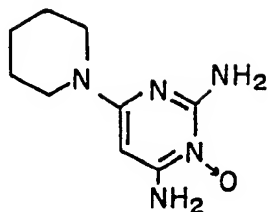
15

20

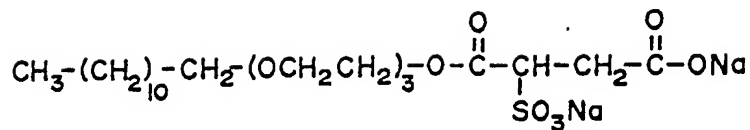
25

30

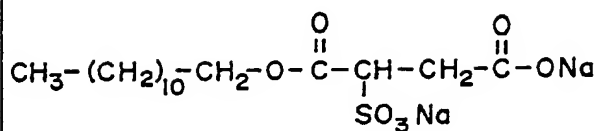
1 / 9



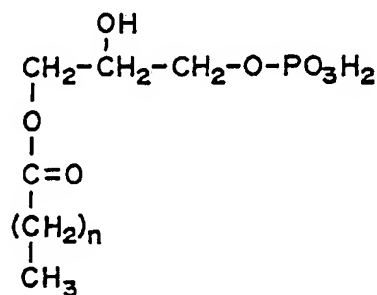
a. MINOXIDIL



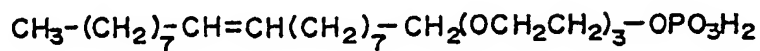
b. LAURETH SULFOSUCCINATE HEMI-ESTER



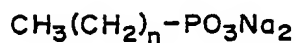
c. LAURYL SULFOSUCCINATE HEMI-ESTER



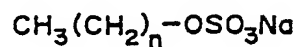
d. LYSO PHOSPHATIDIC ACID



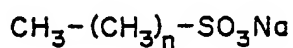
e. CRODAFO3 N3 ACID (OLETH 3 PHOSPHORIC ACID)



f. ALKYL PHOSPHONATE



g. ALKYL SULFATE ESTER



h. ALKYL SULFONATE

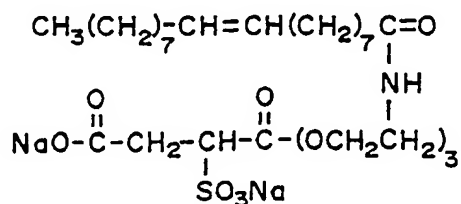
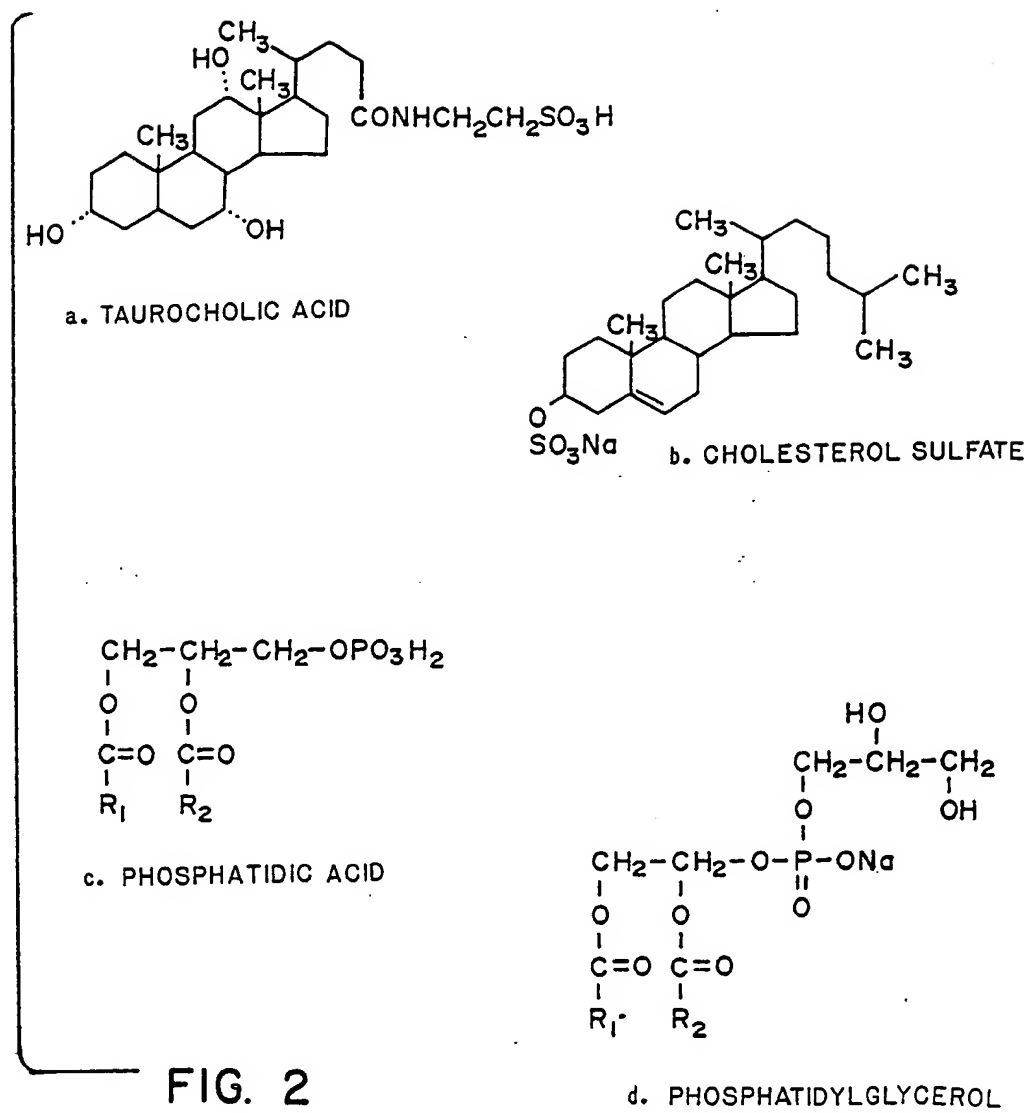
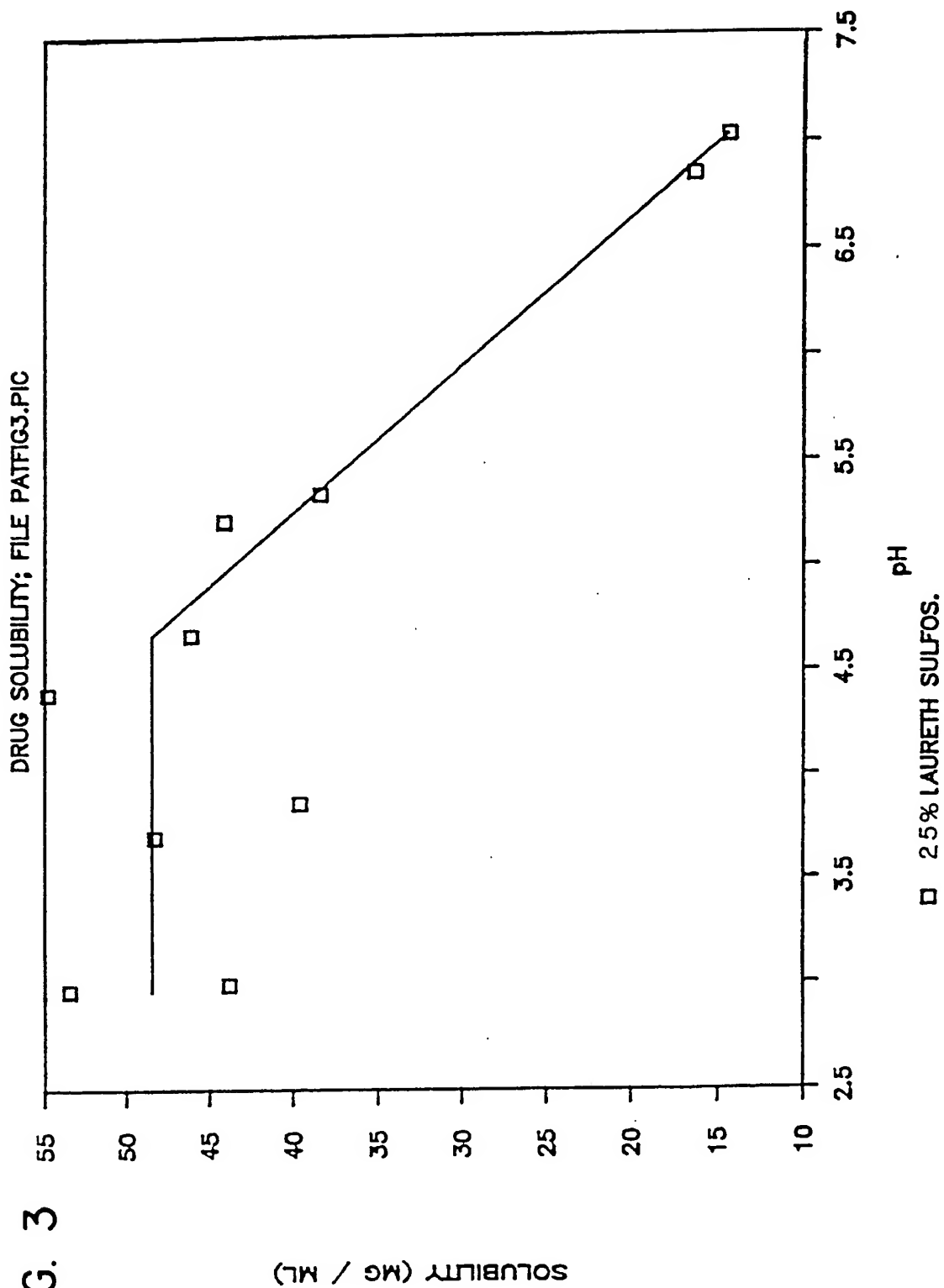
i. OLEAMIDO POLYETHYLENE
i. GYCOL-z SULFOSUCCINATE
HEMI-ESTER

FIG. 1



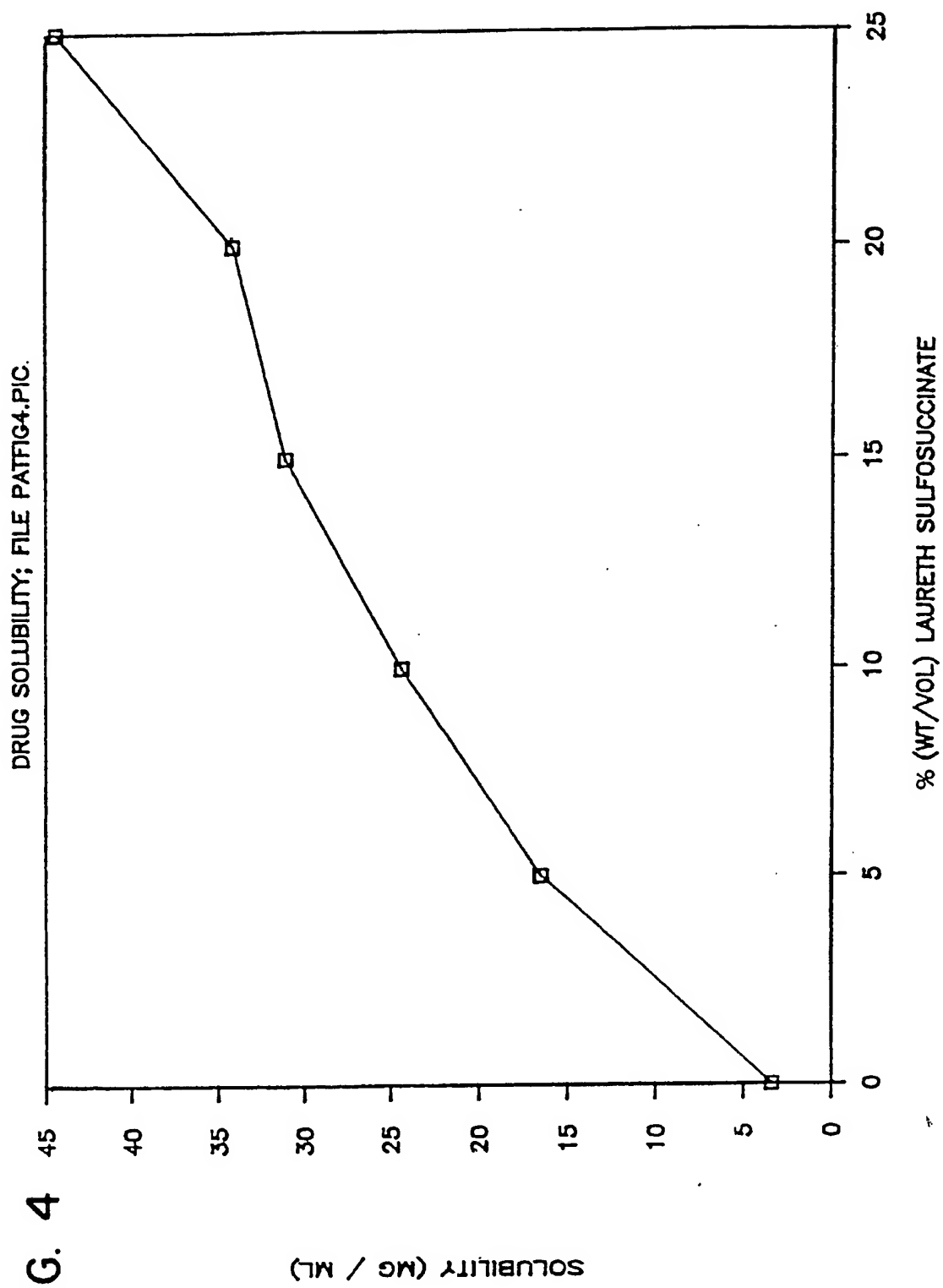
WHERE R_1 AND R_2 REPRESENT ANY FATTY ACID.

3 / 9



SUBSTITUTE SHEET

4 / 9



5 / 9

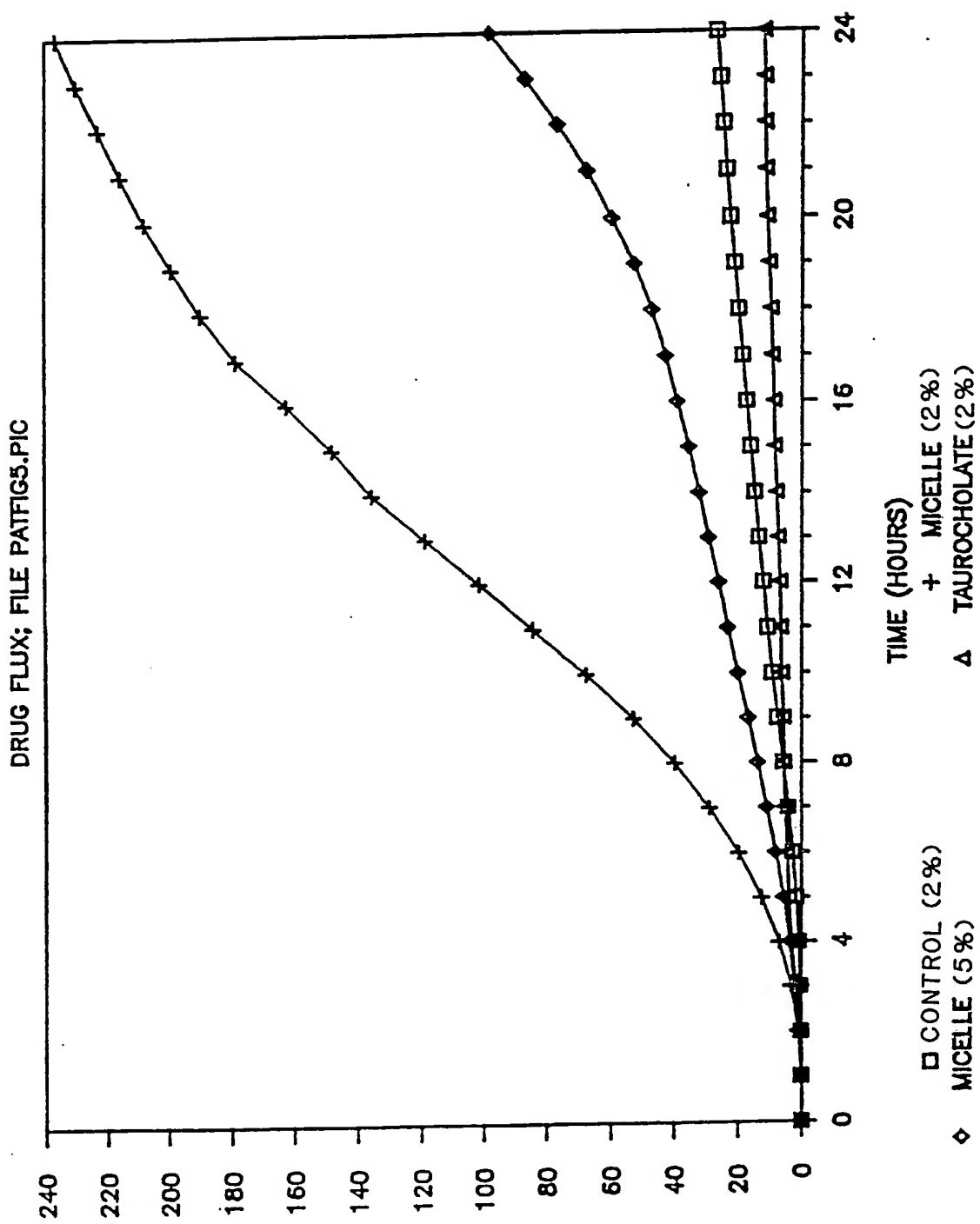


FIG. 5

MICROGRAMS DRUG PENETRATING / SQ CM SKIN

6 / 9

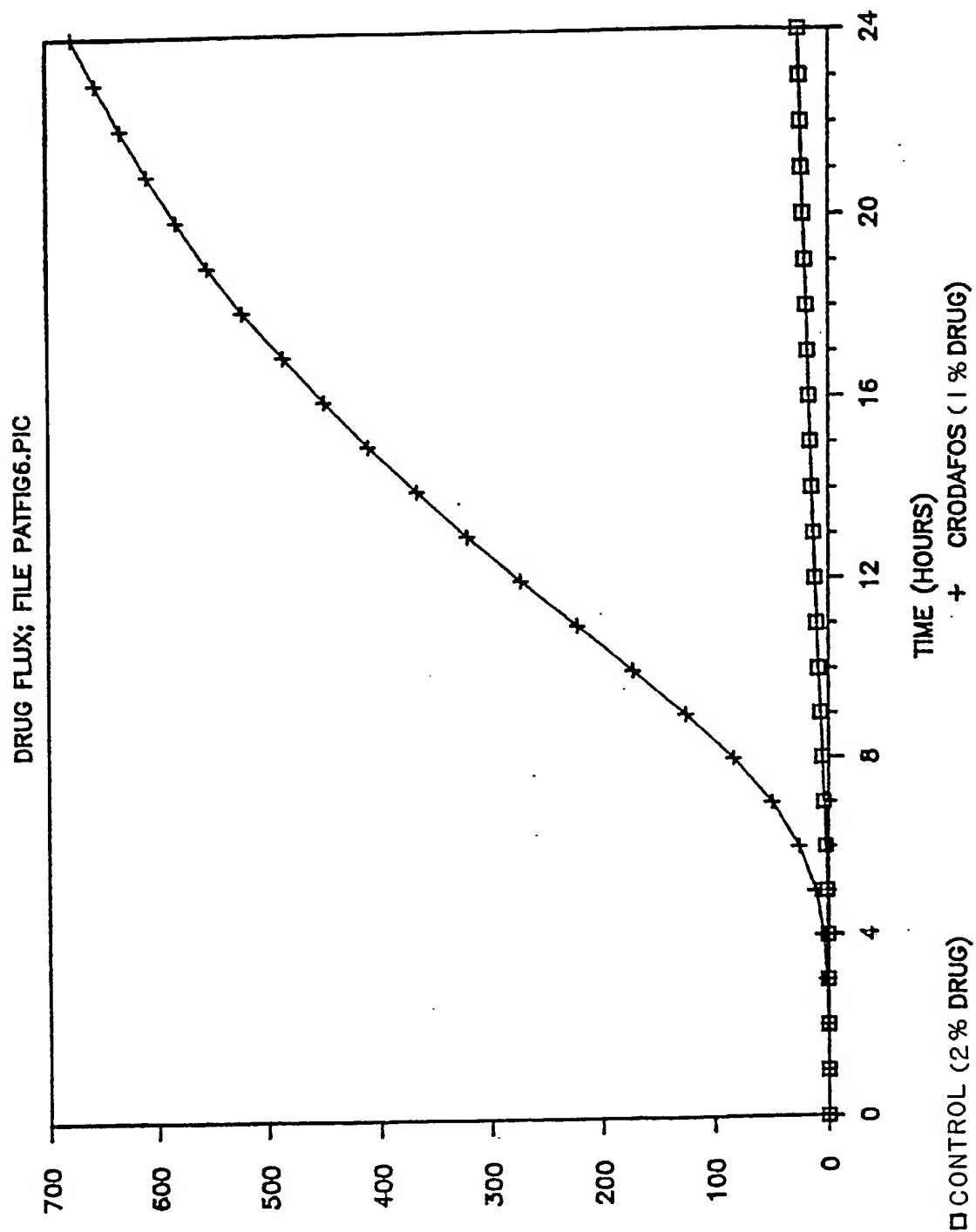
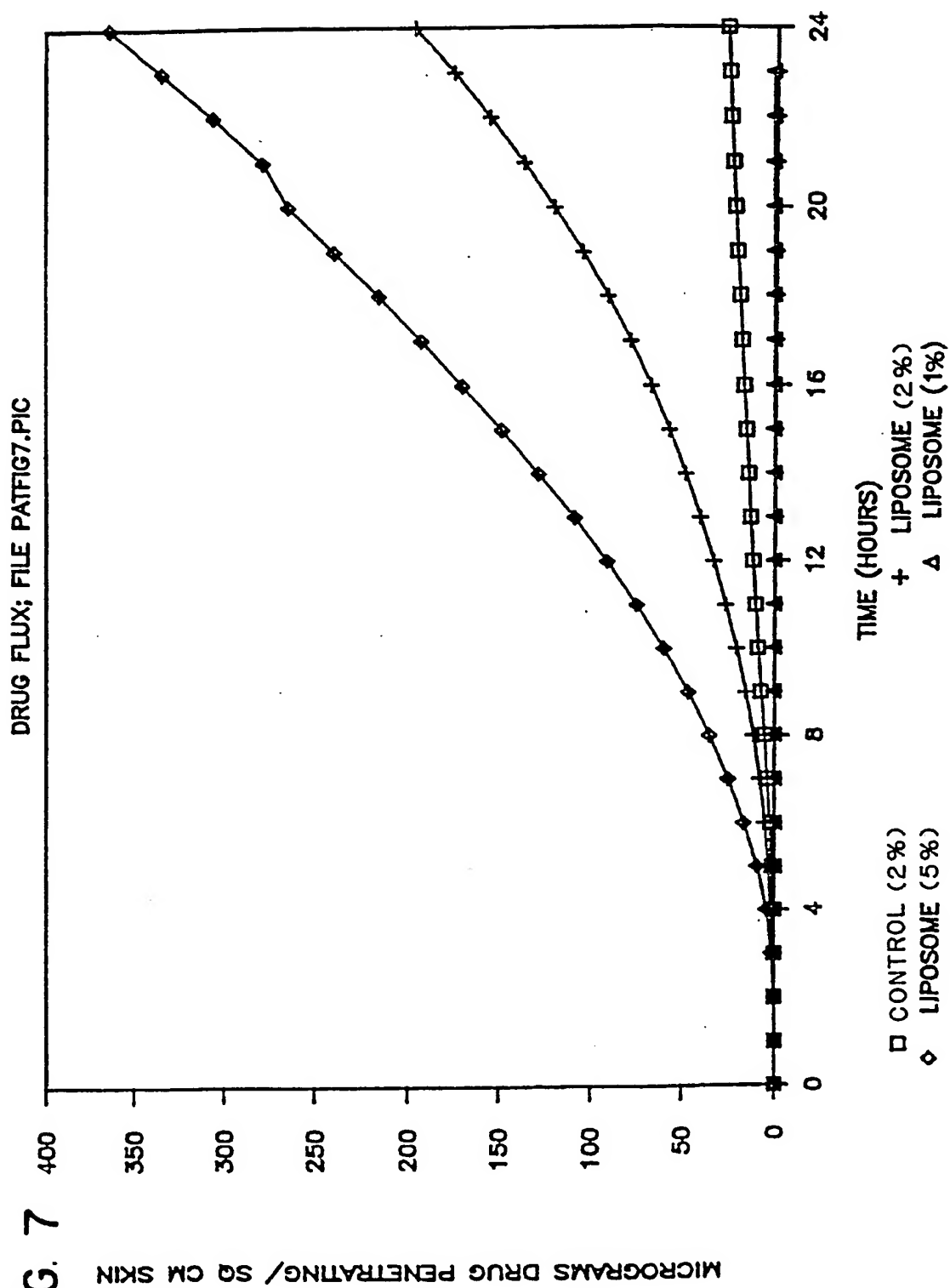


FIG. 6

MICROGRAMS DRUG PENETRATING / SQ CM SKIN

7/9



SUBSTITUTE SHEET

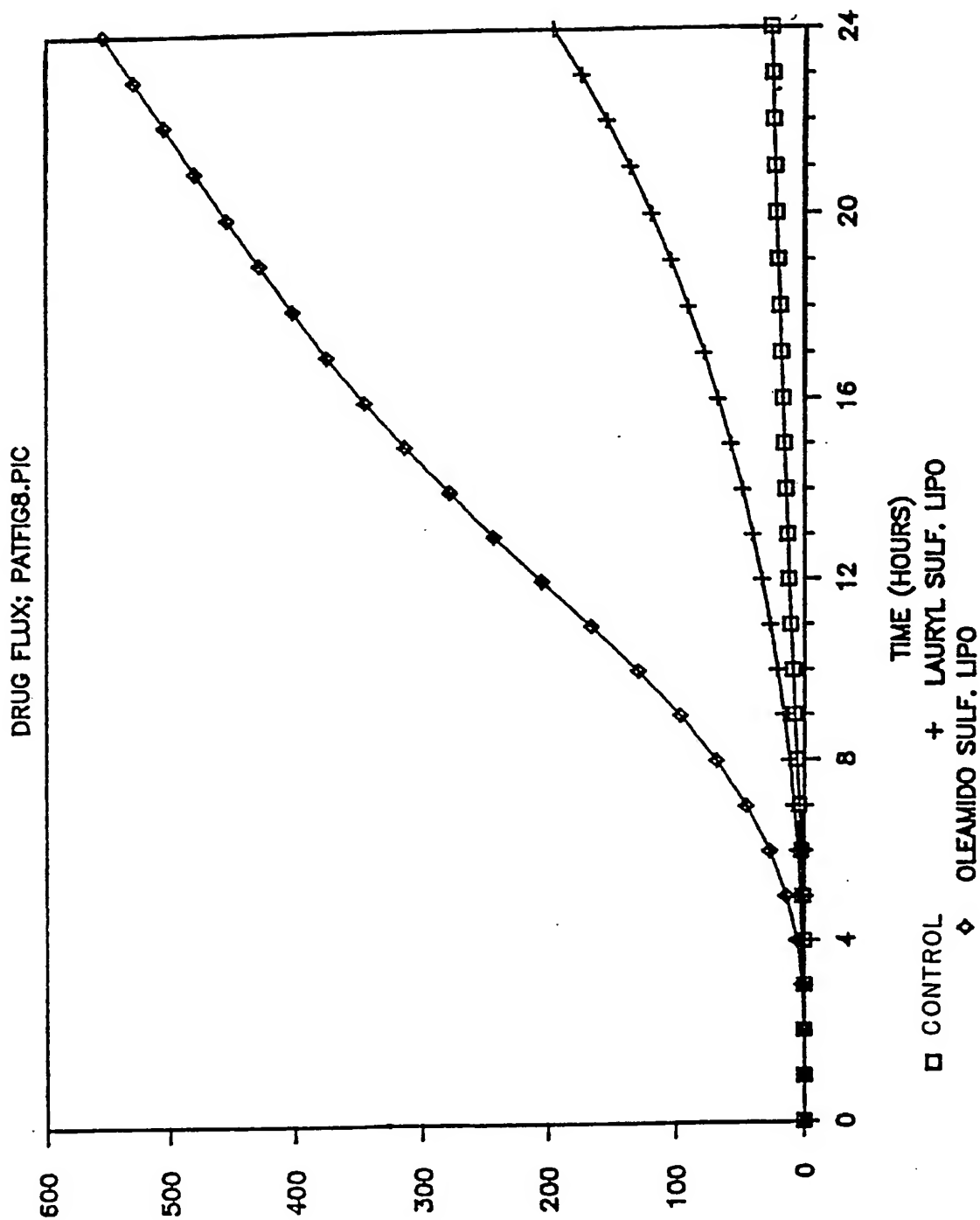
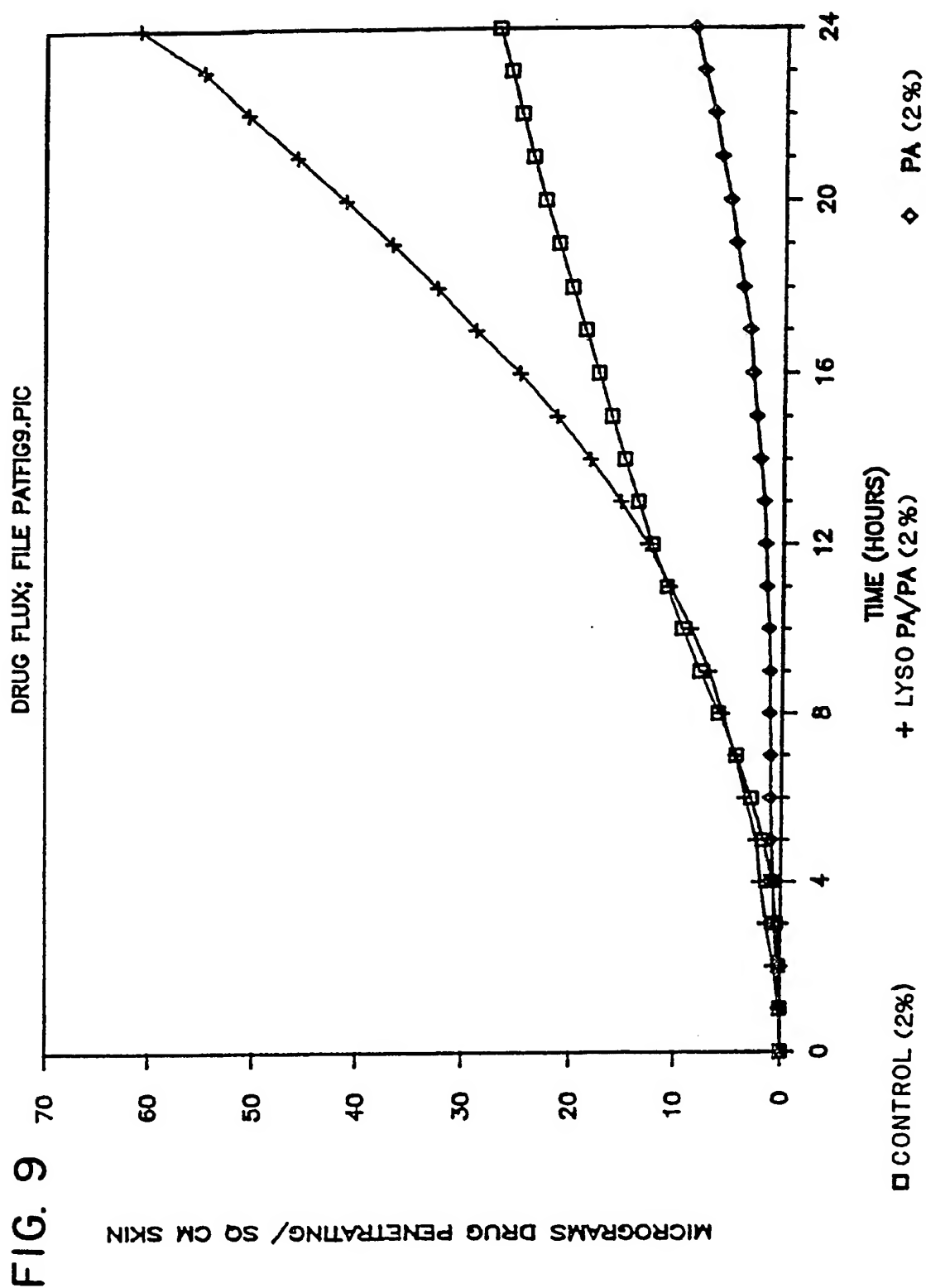


FIG. 8

MICROGRAMS DRUG PENETRATING / SQ CM SKIN

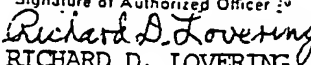
9 / 9



SUBSTITUTE SHEET

INTERNATIONAL SEARCH REPORT

International Application No PCT/US88/00995

I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all) ³		
According to International Patent Classification (IPC) or to both National Classification and IPC INT. CL. 4 A61K 7/06, 9/66, 31/685, 37/22; B01J 13/02. U.S. CL. 264/4.1; 424/45, 450; 428/402.2; 436/829; 514/78, 947, 969.		
II. FIELDS SEARCHED		
Minimum Documentation Searched ⁴		
Classification System ¹	Classification Symbols	
U.S. CL. 264/4.1; 424/45, 450; 428/402.2; 436/829; 514/78, 947, 969.		
Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched ⁵		
III. DOCUMENTS CONSIDERED TO BE RELEVANT ¹⁴		
Category ⁶	Citation of Document, ¹⁵ with indication, where appropriate, of the relevant passages ¹⁷	Relevant to Claim No. ¹⁶
Y	EP, A, 0, 161, 445 PUBLISHED 21 NOVEMBER 1985 GREENX CROSS CORP. SEE ABSTRACT; PAGE 3, LINE 16-PAGE 4, LINE 2; PAGE 7, LINE 14-PAGE 8, LINE 11; AND EXAMPLES.	1-3, 5-19
Y,P	US, A, 4,654, 354 PUBLISHED 31 MARCH 1987 SHROOT ET AL SEE COL. 5, LINES 26-41; COL. 6, LINES 3-9 AND 55-57; AND COL. 7, LINES 5-12.	1-3, 5-19
Y	GB, A, 2, 145, 107 PUBLISHED 20 MARCH 1985 MCGURK ET AL. SEE PAGE 1, LINE 50-PAGE 2, LINE 4; PAGE 2, LINES 18-35 AND 43-58; EX. 1; AND ABSTRACT.	6-8, 11, 15, 17, 18
A,P	US, A 4, 670, 185 PUBLISHED 02 JUNE 1987 FUJIWARA ET AL.	1-19
<div style="display: flex; justify-content: space-between;"> <div style="width: 45%;"> <p>¹³ * Special categories of cited documents:</p> <p>"A" document defining the general state of the art which is not considered to be of particular relevance</p> <p>"E" earlier document but published on or after the international filing date</p> <p>"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)</p> <p>"O" document referring to an oral disclosure, use, exhibition or other means</p> <p>"P" document published prior to the international filing date but later than the priority date claimed</p> </div> <div style="width: 45%;"> <p>"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention</p> <p>"X" document of particular relevance: the claimed invention cannot be considered novel or cannot be considered to involve an inventive step</p> <p>"Y" document of particular relevance: the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.</p> <p>"Z" document member of the same patent family</p> </div> </div>		
IV. CERTIFICATION		
Date of the Actual Completion of the International Search ²		Date of Mailing of this International Search Report ³
06 JUNE 1988		29 JUN 1988
International Searching Authority ¹		Signature of Authorized Officer ²⁰
ISA/US		 RICHARD D. LOVERING